TECHNICAL ABSTRACT

1. AGENT

The agent, hFVIII(V), is a purified formulated retroviral vector preparation encoding only a gene for human factor FVIII (hFVIII) intended for the treatment of severe hemophilia A (congenital FVIII deficiency). The hFVIII gene is a truncated gene and codes for a functional protein. The vector is a Murine Leukemia Virus (MLV) derived system which is produced by an amphotropic packaging line (HAII) derived from a human cell line (HT1080). Expression is driven by the vector Long Terminal Repeat (LTR). The packaging cell line and vector are constructed to minimize regions of homology between vector and packaging components in order to reduce the chance of a recombination event. The producer cell line has been rigorously tested for the accidental production of replication competent retrovirus by standard cocultivation assays at multiple steps, and has been uniformly negative. The producer cell line has been banked and tested according to conventional FDA guidelines for these agents. Cells from the working cell bank were grown at large scale in medium containing fetal calf serum, the supernatant harvested, processed, formulated and stored at –70°C, pending QC testing and release.

2. PRECLINICAL EFFICACY AND TOXICOLOGY

Treatment of normal rabbits and dogs with hFVIII(V) by peripheral vein administration, resulted in therapeutic levels of hFVIII. These levels persisted for as long as two years. Both adult and juvenile animals produced hFVIII following hFVIII(V) treatment. There is evidence of a dose response. and re-administration of hFVIII(V), after hFVIII levels had dropped, increased the level of circulating hFVIII. Hemophilic dogs showed shortening of the whole blood clotting time for variable periods after administration of hFVIII(V). The expected species-specific immune response to hFVIII was demonstrated in these dogs and complicated any observation of clinical efficacy. There were no effects on general health, hematology or clinical chemistry parameters up to 21 months after hFVIII(V) treatment. hFVIII(V) was generally well tolerated in mice up to 26.7×10^8 TU/kg. . Overt toxicity was seen in 2/10 rabbits treated with a dose of 16.7×10^8 TU/kg which is seven times higher than planned for human administration. Doses planned for human administration were not associated with acute or chronic toxicity in mice, dogs or rabbits.

3. BIOLOCALIZATION

PCR analysis to detect vector-specific sequences in the tissues of rabbits treated with hFVIII(V) showed that the spleen and liver were the most highly positive. Vector-specific sequences were seen less frequently in other tissues, including kidney, lung, bone marrow, lymph node and testis. Localization of vector-specific sequences to tissues was not associated with any changes in histopathology. In testis, the low frequency with which vector-specific sequences were detected suggests that they are present near the limit of assay sensitivity (1 in 150,000 cells).

4. CLINICAL STUDY

Phase I is an uncontrolled, open label dose escalation study to establish the safety of intravenous infusions of hFVIII(V) at escalating doses in the range of 2.8×10^7 TU/kg to 2.2×10^8 TU/kg. A second study objective will be to determine whether one or more doses results in the therapeutic target response of at least 7% FVIII sustained over a 12 week period. Each subject will receive a single course of treatment by intravenous administration of equally divided doses on three successive days. The same total dose will be administered to three subjects. These three will be monitored for inhibitor formation and other adverse effects for a period of 4-7 weeks before three additional subjects are treated at the next higher dose. Subjects eligible for Phase I must have a diagnosis of severe hemophilia A (<1% FVIII); be at least 25 years old; be previously treated on at least 100 occasions with FVIII concentrates; have no present or past FVIII inhibitor; and be sterile (due to vasectomy or some other medically documented condition). Subjects who are human immunodeficiency virus (HIV) positive must have CD4 cell counts >300 cells/mm^3 and not be treated with reverse transcriptase inhibitor medication. Subjects who are hepatitis C (HCV) positive must not have clinical or laboratory signs of liver failure. Phase I safety endpoints include adverse clinical events and laboratory tests for: FVIII inhibitor activity; hepatic, renal, hematologic and other organ function; replication competent retrovirus (RCR); CD4 cell count and viral load in HIV-positive subjects; and viral load in HCV-positive subjects. Abnormal laboratory values will be interpreted with reference to underlying chronic conditions. The study will be stopped if any subject develops a positive RCR test, or if more than one subject develops a clinically significant FVIII inhibitor. Efficacy will be assessed by measurement of FVIII activity three times weekly using a one-stage coagulation assay. In addition, FVIII response will be measured by chromogenic assay for FVIII activity, by activated partial thromboplastin time (aPTT), and by ELISA for FVIII antigen. Subjects will record all bleeding episodes, FVIII concentrate treatments, and adverse events on a home diary record. The trial will have two stages: Phase I (approximately 13 weeks); and Phase I-Extension (approximately 40 weeks). Subjects completing the Phase I-Extension will be enrolled in a lifelong surveillance registry for evaluation of long-term safety.